REMARKS

The Final Office Action of December 31, 2002 presents the examination of claims 69-85, 87-107 and 109-125. Applicants filed an Amendment on May 30, 2003, but such Amendment was deemed to raise new issues for consideration by the Examiner and so was not entered.

The present paper cancels all previously pending claims except claims 107 and 109-111 and presents new claims 126-134 for examination.

Claim 107 is amended herein to remove language that merely describes function that results from the otherwise recited structure and further to recite proper Markush language.

Claim 109 is amended herein to make a minor editorial amendment to delete a now unnecessary designation of the promoter sequence as "(I)" and to recite proper Markush language.

Furthermore, the specification is amended at page 15, line 4 to correctly describe the peptide sequence shown there as a portion of SEQ ID NO: 3 rather than the erroneous designation of SEQ ID NO: 1 currently stated. The specification is further amended at page 16, line 2 to recite the correct Sequence Listing identifier for the peptide sequence shown there.

No new matter is introduced by any of the above amendments to the application. Nor does any of the above amendment

introduce any new issue for consideration by the Examiner. Entry of the amendments is requested as the amendments place the application into condition for allowance, or in any event place the application into better condition for appeal.

Support for new claims

New claims 126-127, 130-131 and 133-134 recite peptide sequences for the inducer peptide of the invention. These peptide sequences are found at page 15, lines 3-4 and page 16, lines 1-2 of the specification and in the Sequence Listing.

New claim 128 recites an embodiment of the invention in which the promoter of the invention is operatively linked to a polynucleotide encoding enzymes having particular activities. Support for this embodiment is provided by the specification at page 12, lines 7-10.

New claim 129 recites a vector comprising a promoter of the invention operatively linked to a restriction site. Support for this embodiment is provided at page 5, line 28.

New claim 132 recites a vector comprising a promoter of the invention operably linked to a polynucleotide obtained from a source other than a *Lactobacillus* cell. Support for this embodiment is provided at page 2, lines 17-18, which defines "heterologous expression" as expression of "a gene from a different species". This text should be taken with the text of

the Examples, which exemplify promoters of the invention isolated from different *Lactobacillus* species. Thus, the specification clearly shows that the inventors contemplated embodiments in which promoters exemplified by those of Figure 4 (that is, from *Lactobacillus* species, see page 10, lines 5-14) controlled expression of genes obtained from species other than *Lactobacillus*, that is, "heterologous expression".

Substitute Sequence Listing

A substitute Sequence Listing is also provided that corrects the nucleotide sequences 6-8. The correction is deletion of the last "residue" N, which was inadvertently included. Sequences SEQ ID NOS: 6-8 represent sequences shown in Figure 4. In Figure 4, N designates the beginning of the transcribed portion of the gene following the illustrated promoter sequence (i.e. the beginning of the mRNA) and is not considered part of the promoter sequence.

The printed copy of the revised Sequence Listing is identical to the CRF copy provided on the attached diskette in the file 1380—0122P.ST25.txt, except that the CRF copy lacks formatting information.

Rejection under 35 U.S.C. § 112, first paragraph

Written Description

Claims 69-85, 87-107 and 109-125 stand rejected under 35 U.S.C. § 112, first paragraph, for alleged failure of the specification to provide adequate written description of the invention. Claims 69-85, 87-106 and 112-125 are canceled, rendering the rejection moot as to those claims. This rejection is respectfully traversed as applied to the pending claims. Reconsideration and withdrawal thereof are requested.

The Examiner takes a position that the present application does not adequately describe sufficient species of IF, SakK and SakR genes to support the present generic scope of the claims.

Applicants submit that the instant rejection is now irrelevant to the pending claims 107 and 109-111, as these claims recite none of the IF, SakK and SakR genes, but rather recite limitations of specific nucleotide sequences having a recited spacing. Claims 126-127 are dependent from claim 109 and add a second feature of a peptide having a recited sequence. All of these features are explicitly shown in the Figures of the instant application and in the Sequence Listing and thus these claims should be deemed free of the instant rejection. (Claims 128-134 are similar to claims 109 and 126-127 in all of these regards.)

As to the scope of the promoter, the Examiner has previously conceded that the application discloses the common structural feature of the promoter. (*See*, the previous Office Action, p. 3, last paragraph.) The Examiner now takes a position that the specification only describes the structure of about 20% of the promoter and that the specification does not specify whether the structure that is described is correlated with a function of binding a repressor or an activator. (*See*, the paragraph bridging p. 4-5 of the Office Action.)

Applicants disagree. First, the structural description of the promoter provided in the claims is entirely sufficient to specifically identify a promoter of the invention. As evidence of this, Applicants provide again attached Exhibit D[1] ("Resultats de PATTERN6.doc"), results of a search of the GENBANK database of bacterial genomic sequences conducted under the direction of the Inventor. The search pattern reflects the language of the instant claims describing the claimed promoter. Apparent discrepancies in the "gap" parameter compared to the spacings recited in the claims are due to the way the search program must count gap bases and are not relevant. The search program counts the number of bases between the completely defined motifs, e.g. "motif 1" and "motif 2" (12-18, "motif 1" and "motif 2" are the sequences boxed in Figure 4 and encompass the SEQ ID Nos:

[1] Exhibit D was first provided with Applicants' response of May 30, 2003 that was not entered.

specifically recited in the claims) whereas the claims recite spacing between the repeated sequences within the motifs. In any event, the selection of the gap parameter of 12-18 in the search program corresponds to the recitation of spacing of 17-23 in the claims. The Examiner should note that the search results in "hits" of only the Sakacin gene promoters from *Lactobacillus sake* and Plantaracin gene promoters from *Lactobacillus plantarum*. Thus, the structure of the promoter described in the claims is clearly biologically relevant and defines a recognizable genus.

Second, the Examiner asserts a length of the promoter element of 80 nucleotides without any evidence whatsoever to support her claim that so long a sequence is necessary to the function of the inducible element described in Figure 4 of the present application. The Examiner must accept as true statements made in the specification unless she can present objective, scientifically sound reasons or evidence to doubt them. *In re Marzocchi and Horton*, 169 USPQ 367 (CCPA 1971). The Examiner has produced no evidence or reasoning to support her doubt of Applicants' statements that the minimal promoter structure shown in Figure 4 is functional to bind a regulatory protein, which binding is regulated by the activated R gene product.

Furthermore, Applicants submit that the Examiner will not be able to muster such reasoning or evidence. It is well-known in the art that protein binding sites on DNA can be as small as six nucleotides and furthermore, that the protein interaction sites on many regulatory elements are constituted by short repeated sequences separated by one to two turns of a DNA helix, i.e. 17 to 23 nucleotides as illustrated in Figure 4. In this regard, Applicants also provide again Exhibits E (P.A. Riosen et al., *Mol. Microbiol.* 37:619-628 (2000) and F[2] (P.A. Riosen et al., *Mol. Gen.* Genomics 265:198-206 (2001)). These exhibits provide data from experiments showing protein-DNA interactions in the promoter region of the *Spp* operon (an alternate naming of the present *Sak* operon, Exhibit E) and the *Pln* operon (Exhibit F). In Figure 3 of Exhibit E, gel mobility shift assays are used to show that purified regulatory proteins SppR (=SakR), or PlnC or PlnD, bind to promoter element probes of about 60 nucleotides that encompass the repeat elements (shown as LR and RR in Figure 2) disclosed in the present specification in Figure 4. This establishes i) that the short DNA sequence encompassing the repeats is sufficient for binding of the regulatory proteins and ii) that the PlnC and PlnD proteins are functionally equivalent to the SppR (SakR) protein. In Exhibit F, Figure 3 shows the result of mutation of the consensus repeat sequence

[2] Exhibits E and F were first submitted with Applicants' response of May 30, 2003 that was not entered.

upon binding of PlnC protein. The Examiner will note that the change of the 5' flanking (-1) G residue to A, or of the 3' flanking (+2) G residue to A has no significant effect on protein binding. On the other hand, mutation of residue C3 within the repeat completely abolishes protein binding. Similarly, mutation of residue T6 has a strong effect. Thus, these two residues are implicated as protein contact sites. Figure 6 of Exhibit F illustrates DNAseI footprinting experiments using the Pln operon promoter and PlnC and PlnD as the DNA binding proteins. The protected portions of the probe are two short sequences, each of which covers one of the repeat sequences illustrated in the promoter in Figure 4 of the specification.

The data in Exhibits E and F establish that the functional region of the promoter according to the instant invention is defined by the repeat sequences and their spacing as illustrated in Figure 4 of the instant specification. Thus, the Examiner's argument that the specification does not adequately describe the structure function relationship of the promoter element recited in the claims is entirely without merit.

Also, it is irrelevant that the specification does not choose between release of a repressor or binding of an activator. There is no requirement that the Applicant know the theory by which his invention works. *Micro Motion Inc. v. Exac*

Corp. 16 USPQ2d 1001 (DC NCalif), citing *Diamond Rubber Co. v. Consolidated Rubber Tire Co.*, 220 U.S. 428, 435-36 (1911); *Raytheon Company v. Roper Corporation*, 220 USPQ 592 (Fed. Cir. 1983).

Applicants submit that, as the Examiner has admitted in the first instance, the specification adequately describes the structural elements of a promoter effective in the instant invention that are correlated with the function of regulation of the promoter by the IF-K-R cascade. Accordingly, as all of the pending claims recite the structure of the promoter, they should be found to be of scope adequately supported by the specification and free of the instant rejection.

Enablement

Claims 69-85, 87-107 and 109-125 stand rejected under 35 U.S.C. § 112, first paragraph, for alleged lack of enablement by the specification. Claims 69-85, 87-106 and 112-125 are canceled, rendering this rejection moot as to those claims. This rejection is respectfully traversed as to the pending claims. Reconsideration and withdrawal thereof are requested.

The Examiner argues that the claims define, and the specification describes, only 20% of the promoter sequence that provides for inducibility of the IF-K-R gene cluster and that there is no guidance with respect to the remainder of the

structure as to modification that can be performed with retention of function. The Examiner asserts that the claims encompass promoters of low homology to the sequences stated and does not establish any predictable scheme for modifying nucleotides of the promoter with retention of function.

Applicants first note that the Examiner's beginning premise is incorrect, as has been explained above. The structure of the claimed promoter is explicit in the claims, the Examiner provides no basis for her assertion that any feature of the claimed nucleic acid is a nucleotide sequence longer than that set forth in the claims.

Furthermore, the Examiner's rejection is merely speculation. The Examiner provides no reasoning and no evidence that addition of sequences to either end of the recited structure would interfere with efficacy of the regulatory element. The Examiner provides no evidence that the particular sequence of nucleotides in the region between the repeat sequences in any way affects efficacy of the regulatory element.

Still further, it was well within the skill of the artisan as of the priority date of the instant application to make and test variant promoter sequences, e.g. by site-directed mutation or by deletion analysis using a *Lactobacillus* host to assay inducibility of the promoter using expression of a reporter gene. Such experimentation was typical in the art and expected

to have to be performed in testing variant gene structures for activity. Applicants provide again Exhibit G[3], an excerpt from a textbook published in 1992 (pp. 154-155 of "Recombinant DNA", J.D. Watson et al., c. 1992 by Scientific American Books), fully 4 years prior to the filing date of the instant application, as evidence such assays were well-known in the art. Thus, the skilled artisan can utilize the specification, the knowledge in the art at the time the invention was made, and expected experimentation, to perform the invention throughout its full claimed scope. The specification is thus an enabling disclosure. *In re Wands*, 8 USPQ2d 1400 (Fed. Cir. 1988).

Still further, Applicants do not utilize the term "homology" in the claims; two sequences are recited, having a relation of a recited range of spacing. Applicants submit that the skilled artisan is well-enabled to create a promoter element having these features.

The Examiner further argues, as to claim 109, that the skilled artisan does not know the inducer that is effective for regulating the promoter. Applicants are not certain what is intended by this argument and it is entirely without merit. The specification clearly indicates that the IF peptide of Figure 2 (residues 19-37 thereof) is an example of an effective "inducer" and that the SakR gene product of *L. sake* ultimately acts at the

[3] Exhibit G was first presented with Applicants' Amendment of May 30, 2003 that was not entered.

described promoter. Also, the PlnA gene product from *L. plantarum* C11 is described as an effective inducer and that the *PlnC* and or *PlnD* gene product will also be effective in ultimate action at the described promoter.

Applicants submit that the invention as presently claimed, using the teaching of the specification, the knowledge of the art at the time the invention was made and expected experimentation, can be practiced without undue experimentation throughout the scope of the claims. Thus, the specification is enabling of the claimed invention and the instant rejection should be withdrawn.

Rejection under 35 U.S.C § 112, second paragraph

Claims 69-85, 87-107 and 109-125 stand rejected under 35 U.S.C. § 112, second paragraph, as allegedly being indefinite. This rejection is respectfully traversed. Reconsideration and withdrawal thereof are requested. In particular, the Examiner asserts that the claims recite IF, SakK, SakR, PlnA, PlnB, PlnC, etc. [genes] and it is not clear to the Examiner what molecules are encompassed by these designations. The Examiner further states that the phrase "of a lactic acid bacterium" in some claims is confusing because it may imply that genes not from a lactic acid bacterium are encompassed.

Claims 69-85, 87-106 and 112-125 are canceled, rendering this rejection moot as to those claims. The pending claims 107, 109-111 and 126-134 do not recite the IF, SakK, SakR, PlnA, PlnB, PlnC, etc., genes and so this rejection is obviated as to those claims.

Claims 120-123 stand rejected under 35 U.S.C. § 112, second paragraph as being indefinite for alleged failure to recite an essential element. The Examiner asserts that the missing element is, "at least the SakR gene expression product." Claims 120-123 have been canceled, rendering this rejection moot.

For all of the above reasons, Applicants respectfully submit that all of the present claims define patentable subject matter such that this application should be placed into condition for allowance. If there are minor issues precluding allowance of the application that can be addressed by a telephone discussion, the Examiner may contact the undersigned at the telephone number below, to discuss such matters.

Pursuant to the provisions of 37 C.F.R. 1.17 and 1.136(a), Applicants respectfully petition for a two (2) month extension of time for filing a response in connection with the present application. The required fee of $205.00 is attached hereto.

If necessary, the Commissioner is hereby authorized in this, concurrent, and further replies, to charge payment or credit any overpayment to Deposit Account No. 02-2448 for any additional fee required under 37 C.F.R. 1.16 or under 37 C.F.R. 1.17; particularly, extension of time fees.

Respectfully yours,

BIRCH, STEWART, KOLASCH & BIRCH, LLP

By _____

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Attachments: Substitute Sequence Listing and diskette
Exhibits D-G

DRN/mua